

Research Article

Influence of Pulsed Electric Field Pretreatment on Vacuum Freeze-dried Apples and Process Parameter Optimization

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Abstract: Vacuum freeze-drying can be utilized to obtain high-quality dried products, but its application in the food processing industry is restricted by its relatively high energy consumption. However, this problem can be solved with Pulsed Electric Field (PEF) pre-processing. Using a three-factor quadratic orthogonal regression design, this study assessed the effects of changes in various parameters of PEF pretreatment (pulse intensity, pulse duration and pulse number) on the properties of freeze-dried apple slices (productivity per unit area, flavonoid content and chromatic aberration). In comparison with freeze-dried apple slices that were not exposed to PEF pretreatment, those exposed to PEF showed a 30% increase in productivity per unit area, a decrease of nearly 50% in chromatic aberration and unchanged flavonoid content. Our research provides a theoretical basis for the application of PEF as a pretreatment for freeze-dried foods that can be used to overcome the high energy consumption of the freeze-drying process and improve product quality.

Keywords: Apple slices, PEF, process parameters, pulsed electric field, quality, vacuum freeze-drying

INTRODUCTION

The apple (*Malus domestica*) is a fruit that is commonly consumed across the developed world and provides raw material for many processed foods. Apples are rich in bioactive substances and are an important dietary source of phenols, vitamin C, sugars, some organic acids and minerals. Fresh apples have high water content, making them particularly susceptible to rotting. Dehydration is the most commonly used method for prolonging the shelf-life of apples because it can reduce water content and maintain stability (Krokida and Philippopoulos, 2006). Dehydrated apple slices are crispy, low in fat, high in fiber and rich in vitamins and minerals (Lee and Mattick, 1989).

With growing health awareness, consumers are becoming increasingly concerned with the nutritional value, safety and price of processed foods, in addition to their convenience. All methods of drying fruits and vegetables have limitations that preclude perfect satisfaction of all consumer requirements. Existing commercialized methods for dehydrating apples can negatively affect microstructure, flavor, aroma and

color (Krokida *et al.*, 2000; Krokida and Philippopoulos, 2006; Meisami-asl *et al.*, 2010; Toğrul, 2005). For example, the high temperature associated with hot air drying can negatively affect flavor, color, texture and nutritional value (Goula *et al.*, 2006; Zhang *et al.*, 2005). However, freeze-drying is an alternative dehydration method that can be used to produce high-quality dried products (Genin and Rene, 1995), because it requires very low temperature and pressure. Freeze-drying, also called vacuum freeze-drying, involves drying water-containing material by rapid freezing, causing sublimation of ice in the material into water vapor, which escapes from the product. Freeze-drying consists of 3 stages: Freezing, sublimation drying and adsorption drying. One of the most prominent features of freeze-drying is that it can preserve the original porous structure of processed products, leading to excellent rehydration capacity. Moreover, the minimal thermal and chemical degradation associated with freeze-drying preserves volatile and aromatic components more effectively than other dehydration methods and thus nutrient loss is negligible (Krokida *et al.*, 2000; Nam and Song, 2007). Freeze-drying has been shown to be an ideal dehydration method for a

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wide range of products and it is especially suitable for heat-sensitive materials. However, freeze-drying is very time and energy-consuming due to the complex processes of sublimation and desorption, as well as the required vacuum and refrigeration. To achieve the same final water content in a given product, the cost of freeze-drying is generally 200-500% greater than that of hot air drying (De Beer *et al.*, 2007). Because of the high energy consumption and associated high cost of freeze-drying, freeze-dried foods are relatively expensive, which restricts the application of the process in the food industry.

In general, the dehydration process consumes a significant part of the total energy consumed by commercial producers of dried foods. Therefore, new hybrid dehydration technologies that reduce the energy consumption associated with dehydrating food and preserve food qualities are of significant interest to the food industry. A variety of pretreatment and drying techniques have been studied intensively with the goal of reducing the cost of industrial food dehydration. These studies have shown that non-thermal processing techniques can be used to overcome the limitations of traditional thermal processing methods and satisfy consumer requirements regarding the safety and nutritional value of minimally processed foods (Morales-de la Peña *et al.*, 2012). Pulsed Electric Field (PEF) pretreatment produces little impact on the nutritional value and physical product characteristics of processed foods and it has attracted wide attention due to its ability to satisfy the consumer demand for fresh food in a cost-efficient manner. Moreover, PEF pretreatment has significant potential as a method of preserving liquid foods and studies of the effects of PEF on various juices, mixed drinks and milk have been performed. Recent research indicates that PEF can improve the efficiency of extraction of intracellular metabolites from intact plant tissues, as well as increase dehydration and juice extraction rates from treated materials (Min *et al.*, 2007; Plaza *et al.*, 2011). Studies on the application of PEF as a non-thermal drying technique show that PEF pretreatment can expedite food dehydration, and maintain the color, luster, and nutrient content of dried foods. Our research team found that PEF pretreatment of fruits and vegetables greatly expedited hot-air drying and improved nutritional content, while reducing the energy consumption of the dehydration process (Zhenyu and Yuming, 2009). In addition, we have optimized the pulse parameters for hot-air drying of white radishes after PEF pretreatment. Recent research indicates that PEF pretreatment can expedite the freeze-drying process while maintaining product size and appearance by reducing the particle size of ice crystals in frozen histocytes. PEF pretreatment of apple slices reduces the

energy consumed by the process of vacuum freeze-drying. However, few studies have assessed the quality indices of apple slices following PEF pretreatment/freeze-drying or optimized PEF parameters. In this study, a three-factor quadratic orthogonal design was employed to assess the effects of changes in PEF pretreatment process parameters on productivity per unit area, flavonoid content and chromatic aberration in treated apple slices. The relationship between PEF pretreatment, the properties of vacuum freeze-dried apple slices and process energy consumption was characterized. Finally, PEF parameters were optimized to produce high-quality processed apple slices with relatively low energy consumption. Our findings provide an empirical basis for commercial processing of dried apple products with PEF and guidance for process parameter optimization.

MATERIALS AND METHODS

Materials and equipment: Red Fuji apples grown in Taigu, Jinzhong City, China were used as the experimental materials ($92.08 \pm 0.9528\%$, w/w moisture content). An ECM830 PEF generator and the monitoring system were purchased from the BTX Instrument Division of Harvard Apparatus, Inc. (Holliston, MA, USA). The PEF parameters included pulse intensity (5-3000 V/cm), pulse duration (10-10 000 000 μ s), pulse interval (100-10 000 ms) and pulse number (1-99). A rectangular wave (unipolar) was used. The electrode processing chamber was composed of

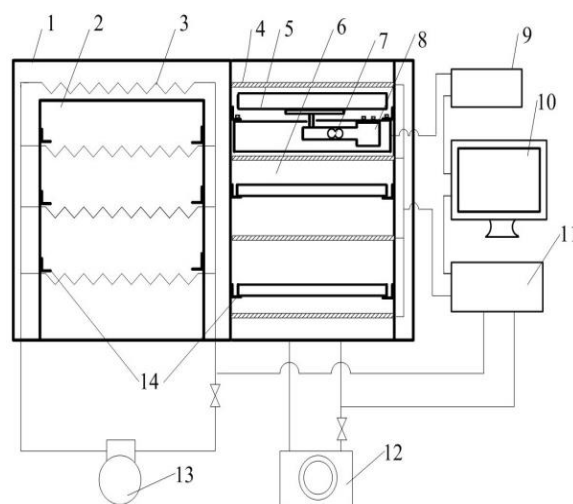


Fig. 1: Schematic diagram of the JDG-0.2 vacuum dryer (A) and the Pulsed Electric Field (PEF) experiment system (B). The system included the following components: (1) dry container; (2) cold trap; (3) condenser tube; (4) electric hot plate; (5) packing box; (6) freeze drying chamber; (7) microcomputer control system; (8) electric device controller; (9) vacuum system; (10) refrigeration system; (11) packing box shelf

two square stainless steel plates (20×20 mm each). The distance between the electrodes was adjustable. A JDG-0.2 vacuum freeze dryer (Lanzhou Kejin Vacuum Freezing Technology Co., Ltd.) was used for the freeze-drying process. The structure and configuration of the equipment are shown in Fig. 1. The drying tests were performed using an online moisture monitoring system designed by Shanxi Agricultural University of China based on the Dynamic Reconfigurable Virtual Instrument (DRVI) and considering the working condition of the JDG-0.2 vacuum freeze dryer (Wu *et al.*, 2011).

The other instruments used in the study included a 7200 spectrophotometer (Unico Instrument Co., Ltd., Shanghai, China), a thermostat water bath (Memmert, Germany), an RE-201D rotary evaporator (Shanghai Yaote Instrument and Equipment Co., Ltd, Shanghai, China) and an SC-80C colorimeter (Beijing Kang Zhuang Instrument Co., Ltd., Beijing, China).

EXPERIMENTAL METHODS

Experimental design and procedure: The selection of pulse parameters and determination of parameter range were based on the performance of the pulse instrument, the bearing capacity of the apple slices and pre-experimental results. Three pulse parameters, pulse strength (10-1500V/cm), pulse duration (50-150 μs) and the number of pulses (1-99), were selected. Pulse interval was not studied due to its minimal influence on the response variables. Our experiments used an orthogonal design with three factors of five levels each to determine the optimal pulse parameters for pretreatment. The following response variables were investigated: Productivity per unit area, flavonoid

content and chromatic aberration. The range and levels of test variables are given in Table 1.

Fresh and intact apples were stored at 4°C. All experiments were finished within one week. Before the experiments, the apples were removed from cold storage. The apples were washed when they reached room temperature, after which surface moisture was blotted to remove it. A rectangle of 20 mm×20 mm×10 mm was cut from the middle of each apple. The apple slices were pretreated by PEF with different combinations of pulse parameters. The initial mass was recorded. Next the freezing temperature of the freezer was set to approximately 243.15K for sample pre-freezing for a period of 2 h.

The drying system was run for at least one hour to achieve stable conditions before sample loading; the preheat temperature was 298.15K. Before the experiments started, the zero point of the data measurement system was adjusted. The dryer was operated when the temperature of the cold trap reached approximately 233.15K. The procedure parameters were set as follows. The temperature of the electrical heating plate was 313.15K during the sublimation process, while the vacuum pressure was 70-75 Pa. The temperature of the electrical heating plate was 343.15K during the desorption process while the vacuum pressure was 25-30 Pa. The parameters were determined based on the relevant literature (Liyang *et al.*, 2012) and early experiments. The data from the virtual experimental platform were stored. Production time and ammeter readings were recorded:

$$P = \frac{M_d}{TS} \tag{1}$$

Table 1: Experimental design and results for treated apple slices (mean ± SD)

Experiment code	X ₁	X ₂	X ₃	Z ₁	Z ₂	Z ₃	Y ₁	Y ₂	Y ₃
1	1	1	1	1282	135	85	123.41±0.62	0.52±0.013	3.5±0.05
2	1	1	-1	1282	135	15	137.44±0.91	0.58±0.008	6.0±0.09
3	1	-1	1	1282	65	85	141.22±0.74	0.60±0.014	7.3±0.12
4	1	-1	-1	1282	65	15	141.58±0.96	0.60±0.007	8.8±0.13
5	-1	1	1	228	135	85	136.55±0.79	0.58±0.005	4.2±0.07
6	-1	1	-1	228	135	15	138.34±0.87	0.58±0.017	5.3±0.10
7	-1	-1	1	228	65	85	141.58±1.02	0.60±0.014	5.8±0.11
8	-1	-1	-1	228	65	15	141.47±0.95	0.60±0.008	6.9±0.10
9	1.414	0	0	1500	100	50	127.83±0.68	0.54±0.015	5.0±0.07
10	-1.414	0	0	10	100	50	141.49±1.13	0.59±0.012	6.3±0.05
11	0	1.414	0	755	150	50	127.67±0.98	0.54±0.004	4.9±0.08
12	0	-1.414	0	755	50	50	141.58±1.21	0.60±0.011	7.9±0.12
13	0	0	1.414	755	100	99	130.21±0.74	0.55±0.014	5.3±0.09
14	0	0	-1.414	755	100	1	138.91±1.09	0.59±0.013	6.8±0.09
15	0	0	0	755	100	50	141.60±1.20	0.60±0.013	4.6±0.11
16	0	0	0	755	100	50	141.58±0.67	0.60±0.015	4.5±0.09
17	0	0	0	755	100	50	141.59±0.96	0.60±0.012	4.6±0.10
18	0	0	0	755	100	50	141.54±1.15	0.60±0.017	4.5±0.08

a) X₁, X₂ and X₃ denote the coding level of pulse strength, pulse duration and number of pulses, respectively; Z₁, Z₂ and Z₃ denote the actual level of pulse strength (V • cm⁻¹), pulse duration (μs) and number of pulses, respectively; Y₁, productivity per unit area g • (h • m²)⁻¹; Y₂, flavonoid content mg • g⁻¹ dry WT; Y₃, chromatic aberration value

where,

P = The productivity per unit area

M_d = The drying weight (g)

T = The drying time (h)

S = The area of the tray (m^2) ($S = 0.36 \times 0.20 m^2$).

Determination of flavonoid content: Flavonoid extraction was performed using heat reflux extraction as reported by Xie *et al.* (2013) and flavonoid content was determined using an $NaNO_2$ -Al (NO_3)₃-NaOH colorimetric method after preliminary experiments.

The dried samples were crushed and 2 g of the sample powder was weighed, added to 65 mL of 60% ethanol and placed into a rotary evaporator. Heat reflux extraction was performed at 70-80°C for 2 h. After the extracts were cooled, they were filtered using a vacuum pump (SHB-III, Greatwall Scientific Industrial and Trade Co. Ltd., Zhengzhou, China). The filtrate was collected and protected from light.

The filtrate (0.5 mL) was mixed well with 0.5 mL of 5% (w/w) $NaNO_2$ in a 10-mL tube. The solution was allowed to stand for 5 min and 0.5 mL of 10% (w/w) $Al(NO_3)_3$ was added. After 6 min, 3 mL of 1 mol/L NaOH was added to the mixture and the resulting solution was brought to a final volume of 10 mL with water. After 15 minutes, the absorbance at 510 nm was measured using a spectrophotometer. For the blank control sample, the same mixture was prepared, but without addition of the sample. The standard curve was plotted with 100 $\mu g/mL$ rutin in 70% ethanol at concentrations from 10-60 $\mu g/mL$ ($y = 0.0868x + 0.00002$, $R^2 = 0.9992$). The concentration of total flavonoids in the extract was expressed as mg of rutin equivalents per gram dry weight of the extract.

Determination of chromatic aberration: Chromatic aberration was determined using the method of Chua *et al.* (2000) with some modifications. The color value of each sample was determined using a chromatic aberration meter (Hunter L, a and b values). The total chromatic aberration was calculated from the chromatic aberration values using the following equation:

$$\Delta E = [(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]^{1/2} \quad (2)$$

where ΔL , Δa and Δb are the differences in the corresponding values of the dried apple slices and fresh apple slices. Lower values of ΔE were associated with less chromatic aberration between the dried and fresh apple slices.

Experiment analysis method: Each treatment was repeated 3 times and all results were expressed as group means. All statistical analysis and modeling were performed using SAS software (SAS Institute Inc., Cary, NC, USA). The significance threshold was set at

$p < 0.05$, with results of $p < 0.0001$ considered extremely significant.

RESULTS AND DISCUSSION

Productivity per unit area: The productivity per unit area of the freeze-dried apple slices that were not pretreated with PEF was 107.11 $g/(h/m^2)$, whereas productivity per unit area was increased in all samples pretreated with PEF (Table 1). The lowest productivity per unit area after PEF treatment was 123.41 $g/(h/m^2)$ and the highest was 141.60 $g/(h/m^2)$, which represented increases of 15.20% to 32.18%, respectively, in comparison with slices not pretreated with PEF.

With the fresh weight of the material and other parameters of freeze-drying kept constant, productivity per unit area mainly depends on the duration of freeze-drying. Therefore, the key factor determining productivity per unit area is the speed of dehydration. The apple slice freeze-drying process consisted of 3 stages. The first stage was sublimation drying, in which the dehydration speed was increased continuously. The second stage was the equilibrium stage, in which the dehydration speed was relatively constant. The third stage was the desorption drying stage, in which little water was removed. The PEF-pretreated apple slices were dehydrated in the first stage of the freeze-drying process much more quickly than were un-pretreated apple slices and the sublimation drying time was reduced by 2 h. The dehydration speed may have been increased because the external electric field applied during the PEF pretreatment caused perforation of the cell membrane, effectively increasing its electrical conductivity and permeability (Toepfl *et al.*, 2005). The small pores created by PEF thus functioned as channels through which water was sublimated and escaped from the cells and freeze-drying energy consumption was decreased.

Regression analysis and significance testing were conducted on the experimental results in Table 1 to obtain a mathematical model of "productivity per unit area". F-tests (Table 2) indicated that the equation was significant, while the lack-of-fit term was not. The experimental data were well fit by the regression model. In addition, the regression coefficients of three indices were determined to assess the influence of the three tested factors on these indices. The parameter with the largest impact on productivity per unit area was pulse duration ($p < 0.01$), followed by pulse intensity ($p < 0.05$) and pulse number ($p < 0.05$). The effects of changes in two parameters are shown in Fig. 2. When the pulse number was fixed (50), the highest productivity per unit area was achieved at a pulse duration of 50-70 μs and a pulse intensity of 500-900 V/cm (Fig. 2a). When the pulse duration was fixed (100 μs), the highest productivity

Table 2: Veriance analysis of the effect of PEF pretreatment parameters on productivity per unit area, flavonoid content and chromatic aberration

	F-value		
	Y ₁ /g · (h · m ²) ⁻¹	Y ₂ /mg·g ⁻¹	Y ₃
Model	10.5 ^b	9.44 ^b	94.840 ^a
X ₁	13.74 ^b	11.767 ^b	41.616 ^b
X ₂	24.30 ^a	21.349 ^a	308.996 ^a
X ₃	8.112 ^b	7.879 ^b	110.915 ^a
X ₁ X ₂	Ns	4.965	50.516 ^b
X ₁ X ₃	Ns	Ns	14.560 ^b
X ₂ X ₃	7.002 ^b	5.674	5.757
X ₁ ²	Ns	Ns	Ns
X ₂ ²	Ns	Ns	Ns
X ₃ ²	Ns	Ns	Ns
Lack of fit	Ns	Ns	Ns
Standard deviation	3.957	0.017	0.653
Mean	141.49	0.59	6.2
Coefficient of variation	4.322	3.119	0.027
R-squared	0.794	0.785	0.896
Adjusted R-squared	0.562	0.543	0.780

X₁, pulse intensity; X₂, pulse duration; X₃, pulse number; Y₁, productivity per unit area; Y₂, flavonoid content; Y₃, chromatic aberration value; ns, non-significant; ^a, p<0.001; ^b, p<0.05

per unit area was achieved at a pulse intensity of 0-500 V/cm and a pulse number of 40-60 (Fig. 2b). When the pulse intensity was fixed (755 V/cm), the highest productivity per unit area was achieved at a pulse number of 5-50 and a pulse duration of 50-60 μs (Fig. 2c):

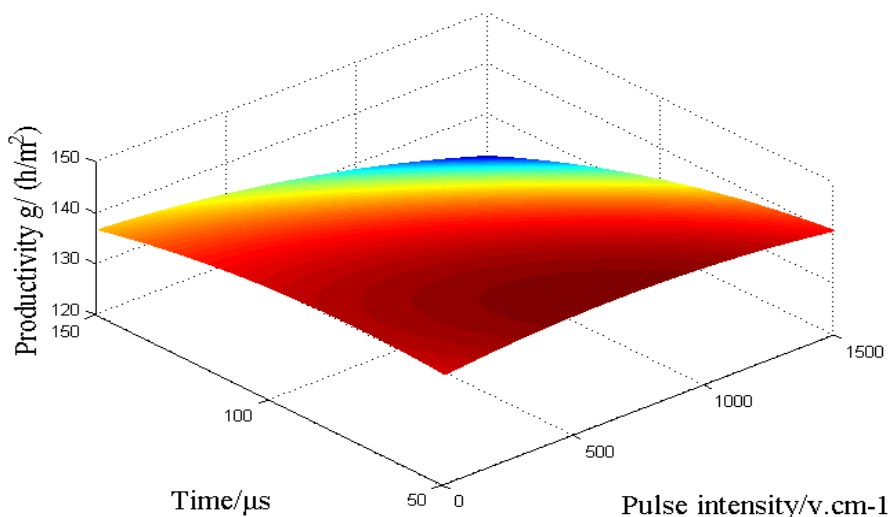
$$\begin{aligned}
 Y_1 = & 125.496518 + 0.014976X_1 \\
 & + 0.241189X_2 + 0.261983X_3 \\
 & - 0.000004501X_1^2 - 0.000092608X_1X_2 \\
 & - 0.001054X_2^2 - 0.000085415X_1X_3 \\
 & - 0.001577X_2X_3 - 0.001078X_3^2
 \end{aligned}
 \tag{3}$$

Flavonoid content: The results of the assessment of flavonoid content are shown in Table 1. The lowest

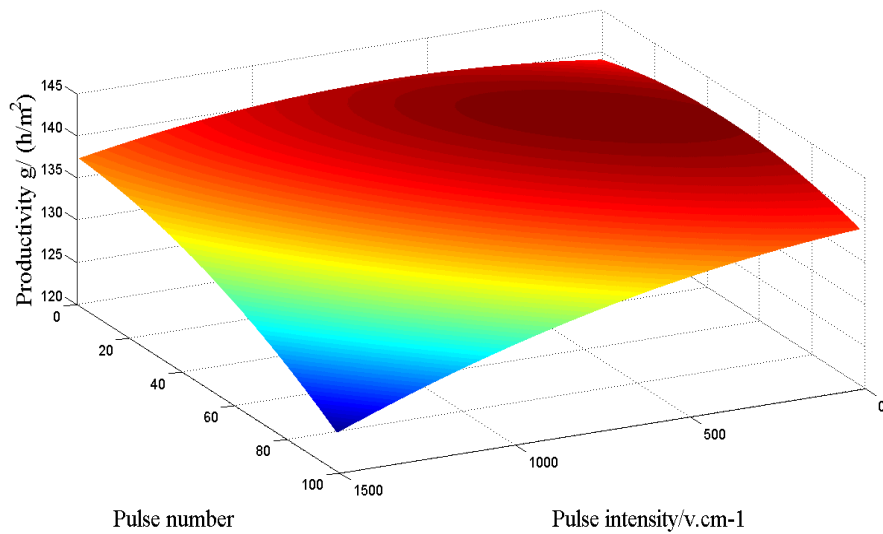
flavonoid content (0.52 mg/g) was measured after PEF treatment with a pulse intensity of 1282 V/cm, a pulse duration of 77 μs and a pulse number of 69. The highest flavonoid content (0.60 mg/g) was measured after PEF treatment with pulse intensities of 1282 V/cm and 755 V/cm, pulse durations of 65 μs and 100 μs and pulse numbers of 15 and 50. The flavonoid content of freeze-dried apple slices that were not subjected to PEF pretreatment was 0.54 mg/g.

Flavonoids are a group of polyphenols that contribute 45%-48% of the total anti-oxidative capacity of apples. Flavonoids lower concentrations of blood lipids and blood pressure, providing a protective effect against cardiovascular and cerebrovascular diseases, as well as tumors, mutations and inflammation (Mejia-Meza *et al.*, 2010; Tsao *et al.*, 2003). A previous study demonstrated that PEF pretreatment did not significantly change the polyphenol content of tomato juice (Odriozola-Serrano *et al.*, 2008). In our study, PEF pretreatment had little impact on flavonoid content, corroborating previous results.

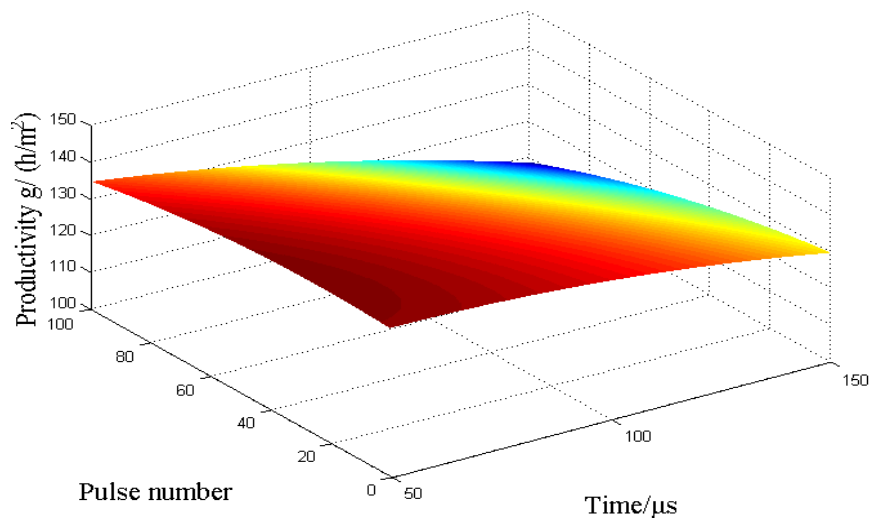
The regression equation for the three tested factors and flavonoid content was obtained. F-tests indicated that the equation was significant, while the lack-of-fit term was not. The experimental data were well fit by the regression model. Of the three tested PEF parameters, pulse duration had the greatest effect on flavonoid content (p<0.0001), followed by pulse intensity (p<0.05) and pulse number (p<0.05). The effects of changes in two parameters are shown in Fig. 3. When the pulse number was kept constant, the highest flavonoid content was achieved at a pulse duration of 50-70 μs and a pulse intensity of 400-900 V/cm (Fig. 3a). When the pulse duration was kept constant, the highest flavonoid content was achieved at a pulse intensity of 200-700 V/cm and a



(a)



(b)



(c)

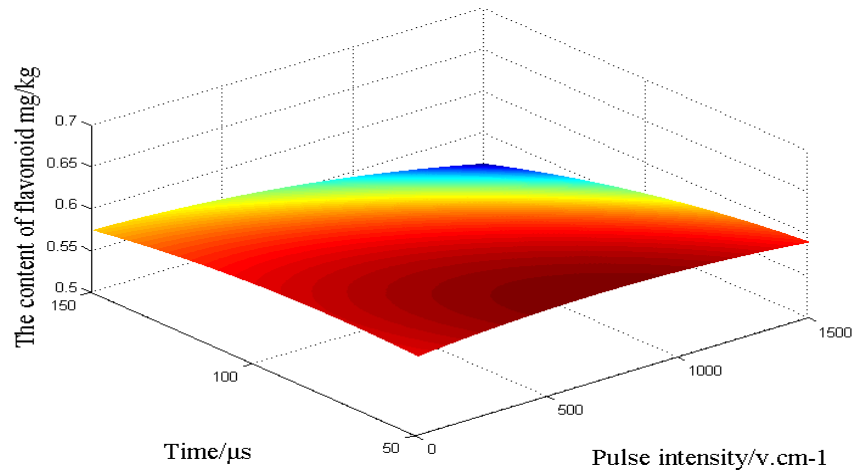
Fig. 2: Effect of changes in two pulse parameters on productivity per unit area of freeze-dried apple slices; (a): Interaction between pulse intensity and pulse duration; (b): interaction between pulse intensity and pulse number; (c): interaction between pulse duration and pulse number

pulse number of 10-60 (Fig. 3b). When the pulse intensity being was kept constant, the highest flavonoid content was achieved at a pulse number of 1-40 and a pulse duration of 50-70 μs (Fig. 3c):

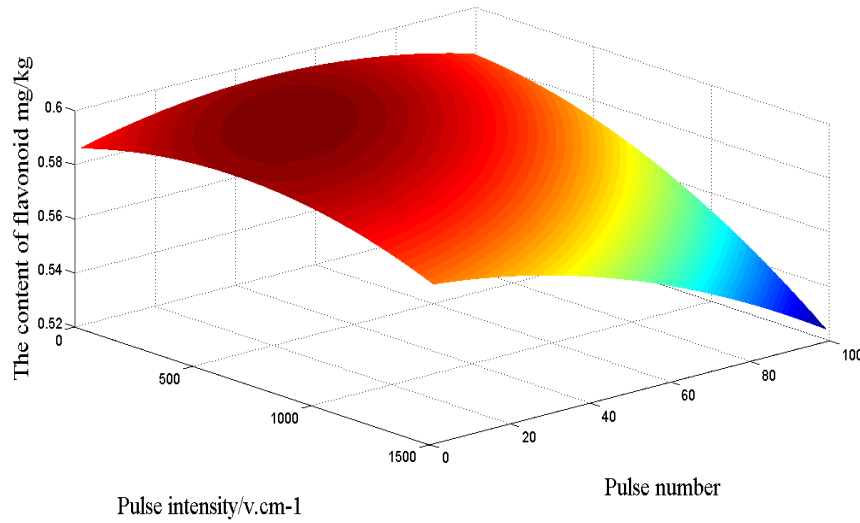
$$\begin{aligned}
 Y_2 = & 0.531989 + 0.000066710X_1 \\
 & + 0.000955X_2 + 0.001027X_3 \\
 & - 0.0000002165163X_1^2 - 0.000000384X_1X_2 \\
 & - 0.000004203X_2^2 - 0.000000352X_1X_3 \\
 & - 0.000006408X_2X_3 - 0.000004129X_3^2
 \end{aligned}
 \tag{4}$$

Chromatic aberration: Chromatic aberration in dehydrated foods, especially in dried food slices, has

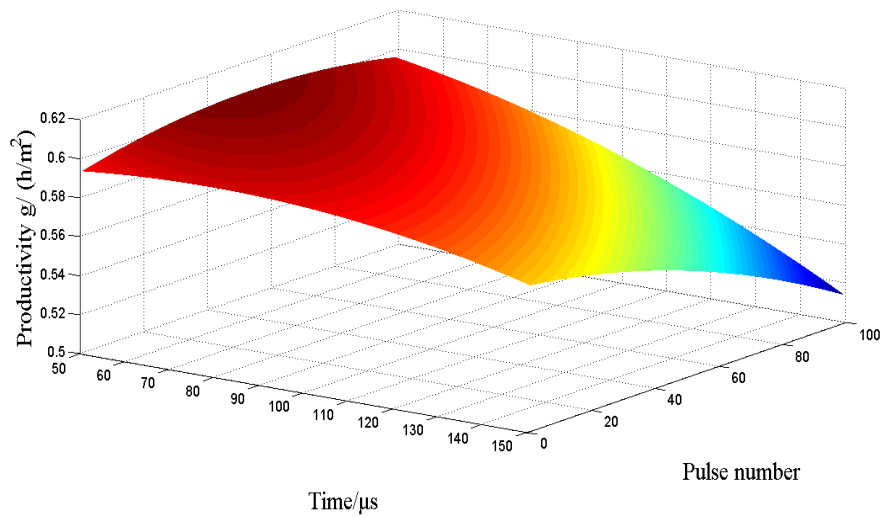
attracted much attention from the food processing industry. In comparison with other fruits and vegetables, apples contain high concentrations of phenolic compounds and enzymatic browning with phenolic compounds as substrates is a serious concern. Polyphenol oxidase (PPO) catalyzes the oxidation of phenolic compounds to form quinones and is the major enzyme involved in enzymatic browning. The action of PPO results in the formation of non-uniform black, brown, or red pigments (Tomás-Barberán and Espín, 2001) that negatively impact the sensory properties of processed foods. Moreover, dietary polyphenols serve as natural anti-oxidants and thus contribute significant nutritional value (Xia *et al.*,



(a)



(b)



(c)

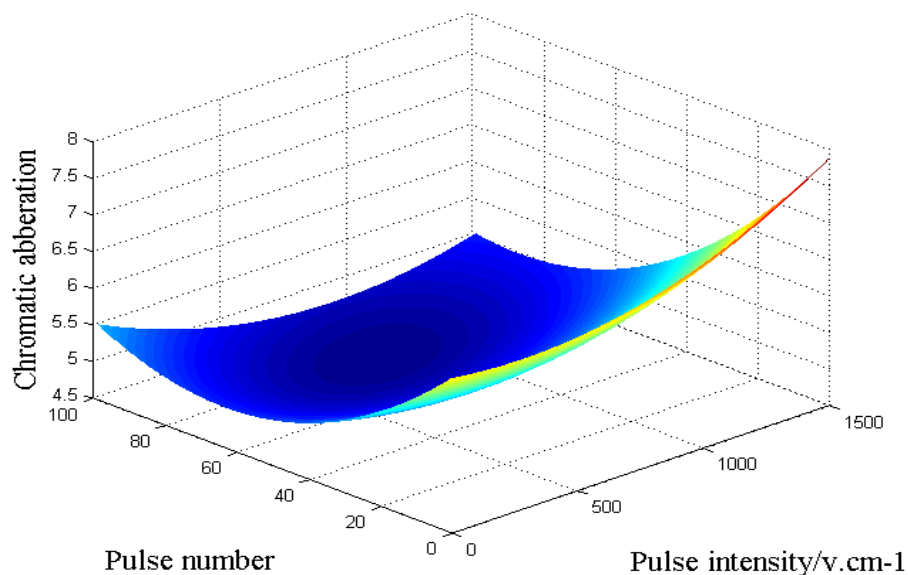
Fig. 3: Effects of changes in two parameters on the flavonoid content of freeze-dried apple slices; (a): Interaction between pulse intensity and pulse duration; (b): interaction between pulse intensity and pulse number; (c): interaction between pulse duration and pulse number

2010). Studies have shown that polyphenols can prevent degenerative diseases, cancer and cardiovascular diseases. Because of its multiple negative impacts on food quality, browning reduces the likelihood that foods are purchased by consumers.

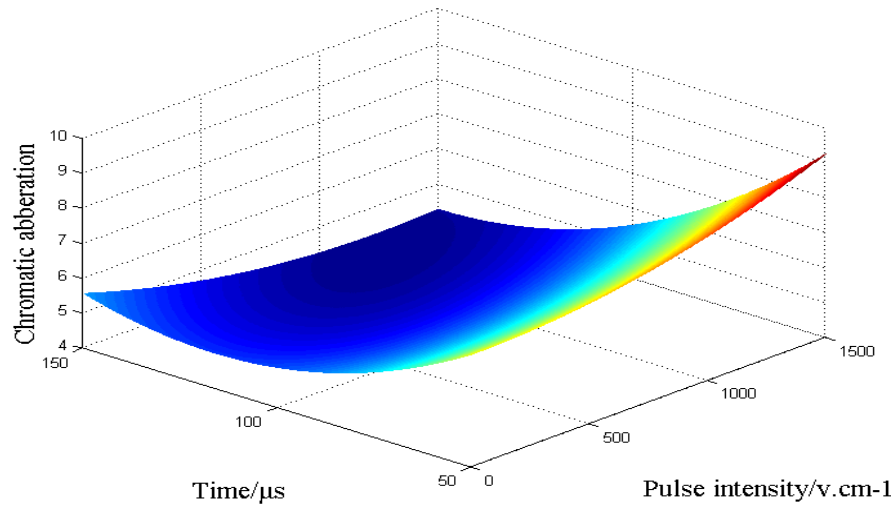
The regression equation for the three tested factors and chromatic aberration was obtained. F-tests showed that the equation was significant, while the lack-of-fit term was not. The experimental data were well fit by the regression model. The chromatic aberration value of the freeze-dried apple slices that were not pretreated with PEF was 8.5. The chromatic aberration value of the pretreated freeze-dried apple slices was increased only by PEF treatment with a pulse intensity of 1282 V/cm, a pulse duration of 65 μ s and a pulse number of 15 and the chromatic aberration value was decreased under all other PEF treatment conditions, with the smallest value being 3.5 (Table 2). The effect of PEF on chromatic aberration may be explained by its influence on enzymes in the treated fruit. PEF can inactivate and enhance the activity of many enzymes (Espachs-Barroso *et al.*, 2003), while it has no impact on some others (Ho *et al.*, 1997). This study determined the PPO activity of fresh apple slices before and after PEF treatment. There was a significant positive correlation between PPO activity and the chromatic aberration value. The PPO activity of pretreated fresh apple slices was 60.03 U/g FW; PPO activity was increased to 69.01 U/g FW by the PEF treatment associated with the highest chromatic aberration value, while PPO activity was decreased to 27.61 U/g FW by the PEF treatment associated with the lowest chromatic aberration value.

The effect of PEF on enzymes depends on the type of enzyme and PEF treatment parameters. After HPEF

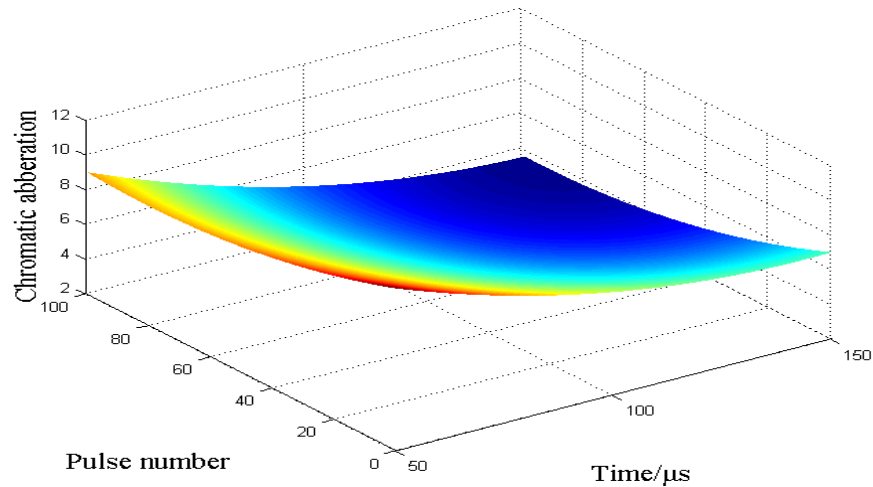
(high pulsed electric field) treatment of milk, fruit and vegetable products, the activity levels of peroxidase (POD), PPO and Hydroperoxide Lyase (HPL) are reduced by 85-100%, while the activity levels of polygalacturonase (PG), lipoxygenase (LOX) and β -glucosidase (β -GLUC) are reduced by less than 50%. Interestingly, a previous study showed that β -GLUC activity was increased to 118.8% of its baseline level by PEF using particular conditions (144.46 Hz unipolar pulse, 1 ms pulse width, 35 kV/cm pulse intensity and 1000 ms pulse time) (Zhao *et al.*, 2012). Similarly, LOX activity in strawberry juice was reduced significantly ($55.8 \pm 1.2\%$) by PEF with a 150 Hz unipolar pulse and a 4 μ s pulse width, whereas LOX activity was increased to $114.1 \pm 1.0\%$ of its baseline level by HPEF with a 250 Hz bipolar pulse and a 7 μ s pulse width (Aguiló-Aguayo *et al.*, 2008). LOX is involved in polyunsaturated fatty acid peroxidation and catalyzes carotenoid cooxidation reactions. High levels of LOX activity result in unpleasant odors and loss of color; therefore, product aroma and color can be preserved and potentially improved, by controlling LOX activity. In this manner, PEF treatment with appropriate conditions can be utilized to maintain optimal levels of enzyme activity and thus produce ideal product color, fragrance and taste. In this study, PEF pretreatment showed an obvious protective effect on the color of freeze-dried apple slices, which was probably due to the fact that the selected pulse parameters inactivated the enzymes involved in browning. Therefore, PEF has the potential to replace chemical treatment with reducing agents for the purpose of color protection in the field of food processing.



(a)



(b)



(c)

Fig. 4: Effects of changes in two pulse parameters on chromatic aberration; (a): Interaction between pulse intensity and pulse duration; (b): interaction between pulse intensity and pulse number; (c): interaction between pulse duration and pulse number

Pulse duration had the greatest influence on chromatic aberration ($p < 0.0001$), followed by pulse number ($p < 0.05$) and pulse intensity ($p < 0.05$). The effects of changes in two PEF parameters on chromatic aberration are shown in Fig. 4. When the pulse number was kept constant, the smallest chromatic aberration value was achieved at a pulse duration of 50-80 μs and a pulse intensity of 0-400 V/cm. The largest chromatic aberration occurred at the highest tested pulse intensity (Fig. 4a). When the pulse duration was kept constant, the smallest chromatic aberration value was achieved at a pulse intensity of 600-900 V/cm and a pulse number of 60-80 (Fig. 4b). When the pulse intensity was kept constant, the smallest chromatic aberration value was achieved at a pulse number of 1-20 and a pulse duration of 50-70 μs (Fig. 4c):

$$\begin{aligned}
 Y_3 = & 12.713201 + 0.001725X_1 \\
 & - 0.110871X_2 - 0.038205X_3 \\
 & - 0.000000892X_1^2 - 0.000022239X_1X_2 \\
 & + 0.000500X_2^2 - 0.000011946X_1X_3 \\
 & - 0.000113X_2X_3 + 0.000387X_3^2
 \end{aligned}
 \tag{5}$$

Process parameter optimization and analysis: The results described above demonstrate that changes in pulse parameters had varying effects on productivity per unit area, flavonoid content and chromatic aberration and changes in some parameters produced both positive and negative effects on the properties of freeze-dried apple slices. For example, both flavonoid content and chromatic aberration values were highest at a pulse intensity of 1282 V/cm, pulse duration of 65 μs

and pulse number of 15. Our results demonstrate that comprehensive optimization using mathematical models that consider quality indicators and processing parameters can be used to achieve high food quality and low energy consumption in food processing applications. Moreover, our findings show that optimization of PEF treatment is crucial for maximizing quality and minimizing cost in industrialized fruit and vegetable processing applications and in the apple industry in particular.

Single objective optimization:

Determination of the objective function:

Optimization was performed for each objective function. The comprehensive scores of productivity per unit area and flavonoid content reached their highest values under the corresponding constraint conditions, while chromatic aberration reached its lowest value:

$$\begin{cases} Y_1 \rightarrow Y_{1\max} \\ Y_2 \rightarrow Y_{2\max} \\ Y_3 \rightarrow Y_{3\min} \end{cases} \quad (6)$$

Constraint condition:

$$\begin{cases} Y_j \geq 0 \\ (i = 1, 2, 3; j = 1, 2, 3) \\ -1.414 \leq X_i \leq 1.414 \end{cases} \quad (7)$$

Optimization results and analysis: The regression model was established, after which the optimization solution was obtained. Optimization was performed with respect to the influence of three pulse parameters on three quality indicators. The optimized productivity per unit area (Y_1) reached a maximum value of 143.72 g/ (h/m²) (coding level: $X_1 = -0.135$, $X_2 = -1.403$, $X_3 = 0.470$; experiment level: $X_1 = 663.289$ V/cm, $X_2 = 51$ μ s, $X_3 = 66.467$). The optimized flavonoid content (Y_2) reached a maximum value of 0.61 mg/kg (coding level: $X_1 = -0.035$, $X_2 = -1.592$, $X_3 = 0.542$; experiment level: $X_1 = 741.518$ V/cm, $X_2 = 50$ μ s, $X_3 = 68.982$). The optimized (minimal) chromatic aberration value (Y_3) was 3.5 (coding level: $X_1 = 1.198$, $X_2 = 1.161$, $X_3 = 0.993$; experiment level: $X_1 = 1386.492$ V/cm, $X_2 = 140.64$ μ s, $X_3 = 74.214$).

Comprehensive optimization of the pulse parameters:

The pulse intensity, pulse duration and pulse number significantly affected the influence of PEF treatment on each quality indicator and every parameter was optimized individually. However, the primary goal of PEF process optimization in this study was to increase the productivity per unit area while decreasing energy consumption and protecting the appearance and quality of the processed apple slices.

Therefore, optimal combinations of different values of pulse intensity, pulse duration and pulse number were determined with respect to their respective influences on quality indicators.

The regression equations for productivity per unit area, flavonoid content and chromatic aberration value were quadratic, non-linear equations; therefore, solving for the optimal parameter combinations was a multi-objective, non-linear programming problem and an evaluation function was needed to convert it into a single-objective programming problem to reach an optimization solution. Greater productivity per unit area, higher flavonoid content and smaller chromatic aberration values were the goals of the optimization. Because of the varying dimensionality of the three objective functions, normalization was first carried out by a linear efficacy coefficient method. The optimal values were calculated by determining the extrema of the regression equations (Xiong and Wang, 2008).

The problem of solving for the maxima of objective functions Y_1 and Y_2 and the minimum of Y_3 was converted into the problem of solving for the maxima of efficacy coefficients k_1 , k_2 and k_3 . The generalized objective function was determined by a linear weighting method:

$$y = k_1 \cdot Y_1 + k_2 \cdot Y_2 + k_3 \cdot Y_3 \quad (8)$$

where k_1 , k_2 and k_3 ($k_1 = 0.5$, $k_2 = 0.2$, $k_3 = 0.3$) were the weighting coefficients of Y_1 , Y_2 and Y_3 , respectively.

A weighted comprehensive optimization was carried out for the optimization results presented above. The optimal parameter combination was determined to include a pulse intensity of 895.890 V/cm, a pulse duration of 77.693 μ s and a pulse number of 69.29; HPEF using these parameters achieved productivity per unit area of 140.42 g/(h/m²), flavonoid content of 0.59 mg/kg and a chromatic aberration value of 4.1. The optimal values of the three parameters were modified to achieve greater experimental feasibility and experimental validation was carried out with a pulse intensity of 895 V/cm, a pulse duration of 77 μ s and a pulse number of 69. Under these parameters, the productivity per unit area was 138.50 g/ (h/m²), the flavonoid content was 0.58 mg/kg and the chromatic aberration value was 4.5. Using these optimized PEF parameters, the productivity per unit area and flavonoid content were similar to the optimal values achieved by single-factor optimization. However, the chromatic aberration value was greater than the lowest value achieved by single-factor optimization, because the main factor influencing chromatic aberration was pulse duration and a small pulse duration led to a large chromatic aberration value.

PEF pretreatment parameter selection and selection of appropriate quality indicator values are important considerations:

Our findings indicate that PEF pretreatment parameter selection and selection of appropriate quality indicator values are important considerations. Some studies have shown that the drying effect of HPEF is mainly dependent on the number and size of the pores in the cell membrane, which is determined by the pulse intensity, pulse number and pulse duration used in the PEF treatment (López *et al.*, 2009); therefore, our assessment of optimal PEF pretreatment was based on the optimal combination of these parameters. Selection of inappropriate parameter values may lead to irreversible enlargement of cell membrane pores or even to destruction of the membrane. As a consequence of these changes in cell membranes, the activity of aquaporins and gap junctions is reduced, which can impair water release and slow the drying process. In this study, the productivity per unit area of freeze-dried apple slices was generally increased by our manipulation of PEF parameters; however, the trends in the changes in flavonoid content and chromatic aberration were different and appropriate pulse parameters were required to prevent decreased flavonoid content and increased chromatic aberration as a result of PEF treatment. When the optimal HPEF parameter combination was used, the productivity per unit area of processed apple slices was increased by 30% in comparison with apple slices not subjected to HPEF pretreatment, while chromatic aberration was decreased by approximately 46%. Our results show that HPEF pretreatment has significant value as a method of reducing the energy consumption and cost associated with freeze-drying fruits and vegetables, while improving product quality.

CONCLUSION

- The influences of PEF pulse intensity, pulse duration and pulse number on productivity per unit area, flavonoid content and chromatic aberration in freeze-dried apple slices were studied. Within the tested range of each parameter, all of the tested pulse parameters significantly influenced the quality of the freeze-dried apple slices and pulse duration had the greatest influence on all three quality indicators.
- Quadratic orthogonal tests were performed to obtain mathematical models of the relationship between each pulse parameter and the three tested quality indicators. The regression coefficients of the model were high and reached significance in the F-test. These results indicate that the model was capable of accurately predicting the selected quality properties within the tested ranges for each parameter.
- Multi-objective non-linear optimization and linear efficacy coefficient methods were employed for comprehensive optimization of PEF pretreatment

of freeze-dried apple slices. The optimal process parameters were as follows: pulse intensity, 895 V/cm; pulse duration, 77 μ s; and pulse number, 69. After PEF pretreatment using the optimal process parameters, the productivity per unit area was 138.5 g/(h/m²), the flavonoid content was 0.58 mg/kg and the chromatic aberration value was 4.5. Moreover, utilization of the optimal process parameters expedited dehydration and processing reduced energy consumption, and improved the quality of the processed freeze-dried apple slices.

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